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# Evaluation of the prebiotic GroBiotic®-A and brewers yeast as dietary supplements for sub-adult hybrid striped bass (*Morone chrysops*×*M. saxatilis*) challenged in situ with *Mycobacterium marinum*

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**Abstract**

Dietary supplements such as immunostimulants and prebiotics hold promise as a potential replacement of antibiotics in maintaining fish health. A 21-week feeding trial was conducted to evaluate the commercial prebiotic GroBiotic®-A, a mixture of partially autolyzed brewers yeast, dairy ingredient components and dried fermentation products, in the diet of hybrid striped bass exposed to chronic mycobacterial infection caused by *Mycobacterium marinum*, as compared to partially autolyzed brewers yeast (Brewtech®). The basal diet was formulated to contain 40% protein, 10% lipid and an estimated digestible energy level of 3.5 kcal/g. Supplements of 1 or 2% brewers yeast and 2% GroBiotic®-A were singularly added to the basal diet and each diet was manufactured by extrusion processing with a twin-screw extruder. Each diet was fed to three replicate groups of small (initially averaging 64.5 g/fish) and one group of large (initially averaging 118 g/fish) hybrid striped bass in 1187-l circular tanks operated as a recirculating system. Fish were fed twice daily to apparent satiation and growth performance monitored for 16 weeks. An in situ infection of *M. marinum* became well established at week 16 such that fish were fed once daily and mortality was monitored for a total of 21 weeks.

Enhanced growth performance was generally observed in fish fed diets supplemented with GroBiotic®-A or brewers yeast compared to fish fed the basal diet throughout the feeding trial with significantly ($P<0.05$) enhanced weight gain observed after 12 weeks of feeding. At the end of the feeding trial, fish fed 2% brewers yeast had significantly higher feed efficiency than fish fed the other diets. The in situ mycobacterial challenge employed in this experiment resulted in overall cumulative mortality of approximately 25%. Fish fed 2% GroBiotic®-A had a significantly ($P<0.05$) enhanced survival (80%) compared to the other treatments (72–73%) at the end of 21 weeks. It is concluded that dietary supplementation of 2% GroBiotic®-A showed moderate but significant ($P<0.05$) protection against mycobacterial infection. Dietary

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supplementation of partially autolyzed brewers yeast also may enhance growth performance under chronic infection of mycobacteria.


*Keywords:* Prebiotics; Hybrid striped bass; GroBiotic®-A; Brewers yeast; *Saccharomyces cerevisiae*; Immune response; Bacterial resistance; Mycobacteria

## 1. Introduction

Rapid growth and disease resistance of aquacultured organisms are two of the most important concerns. Traditionally, antibiotics have been supplemented in aquafeeds for prevention and/or treatment of bacterial disease of aquatic animals. It has been reported that antibiotics may enhance growth and feed efficiency by killing intestinal microflora and thus increasing amino acid utilization by the host in some animal species (Rawles et al., 1997). However, the use of antibiotics may pose threats such as development of bacterial strains that are more resistant to antibiotic treatment, or the occurrence of antibiotic residues in cultured organisms for human consumers (FAO, 2002). Increasing concerns of antibiotic use have resulted in a ban on subtherapeutic antibiotic usage in Europe and the potential for a ban in the United States and other countries (Patterson and Burkholder, 2003). These alterations in policy may impact aquaculture and therefore prompt interest in developing alternative strategies for disease control. Beside vaccine development, dietary supplements including probiotics, prebiotics and immunostimulants have received heightened attention. A rapidly expanding body of literature has been established that many intestinal microbial species may have beneficial influences on the performance of fish (reviewed by Irianto and Austin, 2002), and dietary composition is capable of influencing the intestinal microflora of fishes (Ringø et al., 1998; Ringø and Olsen, 1999). However, development of prebiotics, classified as "nondigestible food ingredients that benefically affect the host by stimulating growth and/or activity of a limited number of bacteria in the intestine", is in its infancy with fishes, compared to the progress that has been made in development of prebiotics for poultry (Patterson and Burkholder, 2003). In a previous evaluation of the commercial prebiotic, GroBiotic®-A, a mixture of partially autolyzed brewers yeast, dairy ingredient components and dried fermentation products, significantly enhanced feed efficiency of juvenile hybrid striped bass was observed (Li and Gatlin, 2004), although the dynamics of the intestinal microflora was not defined in that study. Supplementation of this prebiotic also enhanced respiratory burst of head kidney leucocytes and resistance against *Streptococcus iniae* infection; however, the interpretation of these beneficial influences was complicated by the presence of brewers yeast, which is generally considered to be an immunostimulant for fishes (Siwicki et al., 1994; Ortuño et al., 2002; Li and Gatlin, 2003; Rodríguez et al., 2003).

The hybrid striped bass is an important fish for U.S. aquaculture, but it is affected by several pathogenic bacteria such as *S. iniae*, *Aeromonas hydrophila* and *Mycobacterium marinum* (reviewed by Plumb, 1997). With limited availability of approved therapeutic compounds and the inconvenient and costly administration of vaccines, this fish has been used in our laboratory as a model for investigating the interaction between nutrition and disease resistance. Besides *S. iniae,* the prevalence of mycobacteria in wild striped bass and cultured hybrid striped bass has attracted increased attention (Gauthier et al., 2003; Overton et al., 2003). Although mycobacteriosis is generally chronic, this disease may cause severe infection and high cumulative mortality in closed, recirculating systems (Plumb, 1997). In addition, this bacterium is reported to be capable of surviving under many adverse environmental conditions, including low temperature, and may cause infection on the extremities of humans (Plumb, 1997; Mediel et al., 2003). A recent report of the presence of mycobacteria in frozen seafood also raised concern about the safety of such seafood for human consumers (Mediel et al., 2003). Currently, effective treatment of this disease is very limited as reported for other fish species (Colorni et al., 1998). Therefore, the present study was conducted to explore growth performance